KINETICS OF THE FORMALDEHYDE DETOXIFICATION PATHWAY. <u>C. D. Koch</u>, M.R. Fry\* Bradley University, Department of Chemistry and Biochemistry, Peoria, IL 61625 mfry@bumail.bradley.edu

FSF1 is one of several fusion genes encoding bifunctional fusion proteins that have been identified in ciliates, including Tetrahymena thermophila (TH). The bifunctional protein encoded by FSF1 exhibits both formaldehyde dehydrogenase and S-formylglutathione synthetase activities of the formaldehyde detoxification pathway. Most organisms have two separate genes that encode two distinct enzymes for this pathway. The biological reason for having this fusion gene is not known at this time. In this study, the kinetics of the bifunctional TH enzyme is being compared to the kinetics of the discrete enzymes from Escherichia coli (E. coli). Kinetics of the reverse of the formaldehyde detoxification pathway were followed by monitoring the absorbance change at 340 nm that accompanies the oxidation of NADH to NAD<sup>1</sup>. In this manner, the combined activities of both enzymes was detected. Initial studies showed an increased rate of NADH oxidation for the crude extract of *TH* compared to the crude extract of *E. coli*. We have attempted to partially purify the enzymes from both species using affinity chromatography and have compared activities in the fractions obtained. Enzyme purification techniques are being optimized at this time. To date, our data suggests that the bifunctional enzyme metabolizes formaldehyde at a faster rate than the discrete forms. Substrate channeling of the fused TH enzymes is proposed as a possible explanation for this observation.